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MODIFICATION OF THE VACCINE RESPONSE IN RABBITS BY THE APPLICATION OF DIPHTHERIA TOXIN TO THE VACCINATION SITE

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Various substances have been reported from time to time as exerting a local modifying influence upon the character of vaccination "takes" in animals. Ledingham (1927) reported that India ink exerted a deterrent local influence upon cutaneous and more especially upon intracutaneous vaccinations in rabbits when incorporated with the vaccine virus.

Carnot and his coworkers (1926), Le Fevre (1927), and Rivers and his associates (1928) showed that various forms of irradiation of the skin of rabbits would induce a local refractory state to a subsequently performed cutaneous vaccination. Seiffert (1931) reported that the local response engendered by the application of the appropriate antigen to the skin of sensitized animals will counteract the effect of a weakly virulent vaccine virus applied at the reacting site.

The writer has investigated the influence of various substances incorporated with vaccine virus upon the "take" in rabbits and has found that diphtheria toxin (guinea pig M. L. D. 0.0045 cc) when diluted 1:50 in saline and mixed with an equal volume of vaccine virus exerted a deterrent effect upon both the local and systemic vaccine response.

This effect was noted with various strains of vaccine, but was most apparent with virus highly potent for the rabbit. Consequently, most of our work has been done with the virulent strain of virus previously described (Armstrong, 1929).

The deterrent effect of diphtheria toxin was apparent following varied methods of vaccination but seemed to be most apparent when a superficial type of insertion and a relatively low dilution of toxin were used.

We have, therefore, usually employed an insertion one-half inch in diameter which was made by superficially pricking the dermis, through 0.1 cc of the virus-toxin mixture, by means of a sharp needle. For control purposes a similar procedure was employed except

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that the virus was diluted with an equal volume of diphtheria toxin (1:50 in saline) which had been previously heated for 15 minutes at 98° C.

CHARACTER OF THE "TAKES"

In animals vaccinated with virus plus active diphtheria toxin the lesions at the end of 24 hours consisted of pink, slightly edematous areas, usually about 1 inch in diameter. At the end of two days the lesions were wider and brighter in color, and the central pricked areas were either mottled with purplish subepithelial hemorrhages or, when these were absent, they showed a yellowish cast through the injured epidermis. This central area was often surrounded by a zone of pale yellowish pink bordered on its outer edge by a narrower zone of bright pink which faded into normal skin. More or less maceration of the epidermis was usually present. By the end of 72 hours the lesions had usually begun to fade and shrink, the edema gradually disappeared, and a dry scab with scaling edges resulted. The lesions as described were similar to those produced by diphtheria toxin alone when applied by this method. Often no reaction suggestive of a vaccine "take" occurred. In some instances, however, vaccine lesions evinced by varying degrees of erythema and subcutaneous edema did develop, oftenest at the ventral border of the scab. A few secondary vaccine papules on the epilated areas outside the site of the "Schick" reaction were common. Upon recovery, the animals were immune to vaccinia.

In the group vaccinated with virus plus heated toxin, the lesions at the end of 24 hours were pale pink, slightly elevated spots approximately the size of the original insertion. The following day there was not much change, but by the third day the pink areas were beginning to enlarge and considerable subcutaneous edema was usually present, especially ventrally. The pricked areas at this time usually showed some superficial necrosis with a purplish discoloration. During the next 4 or 5 days the subcutaneous edema usually increased markedly and often became massive along the whole abdomen. The central purple, necrotic areas enlarged also and ultimately dried to thick black scabs. Secondary papules on the epilated areas were common. In order to rule out variations in the susceptibility of different rabbits to vaccinia, several tests were made by vaccinating the same rabbit with both the test and control mixtures. The results showed the same differences as those described.

TABLE 1.—(*a*) *The temperature response (°C) in rabbits vaccinated with vaccine plus raw diphtheria toxin*

Rabbit No.	Days after vaccination										
	0	1	2	3	4	5	6	7	8	9	10
1579	39.0–39.5	39.5–40.0	39.5–	39.9–	40.6–40.6	40.1–41.0	40.9–41.4	40.4–40.5	39.7–39.4	39.0	
1581	39.0–39.6	39.9–39.7	39.6–	40.0–	39.9–40.0	40.7–40.8	40.2–40.0	40.2–40.2	40.2–40.2	39.7	
1583	39.3–39.4	39.4–39.6	39.8–	39.4–	39.7–40.0	39.9–40.0	39.8–40.0	39.6–39.9	39.3–39.3	39.3	
1609	39.5–39.9	39.5–39.5	39.6–39.7	–40.1	40.4–41.0	41.0–41.2	41.5–41.1	41.0–41.0	41.0–41.0	40.5	39.8–39.5
1611	39.0–39.8	39.3–39.6	39.5–39.6	–39.7	40.0–40.9	41.0–40.6	40.6–40.4	39.8–39.6	39.3–39.6	39.1	39.7–39.4
1613	39.5–39.5	39.3–39.7	39.9–39.8	–40.0	39.6–41.0	40.7–40.5	40.8–40.5	40.3–40.0	40.0–40.5	39.7	39.6–39.3
1615	39.2–39.9	39.5–39.8	39.8–39.7	–39.9	39.8–41.1	41.0–40.4	40.5–40.1	39.8–39.9	39.7–40.0	39.3	39.6–39.4
1617	39.2–39.5	39.3–39.9	39.8–39.8	–40.0	39.9–40.5	40.6–39.5	40.0–39.8	39.5–39.5	39.4–39.5	39.6	39.5–39.3
1619	39.3–40.0	39.4–39.8	39.8–40.1	–40.7	40.2–40.3	40.9–40.7	40.6–40.8	40.0–40.1	39.8–40.1	39.5	39.5–39.3
Average temperature	39.2–39.7	39.5–39.7	39.7–39.8	39.8–40.1	40.0–40.6	40.7–40.5	40.5–40.5	40.1–40.1	39.8–40.0	39.5	39.6–39.4

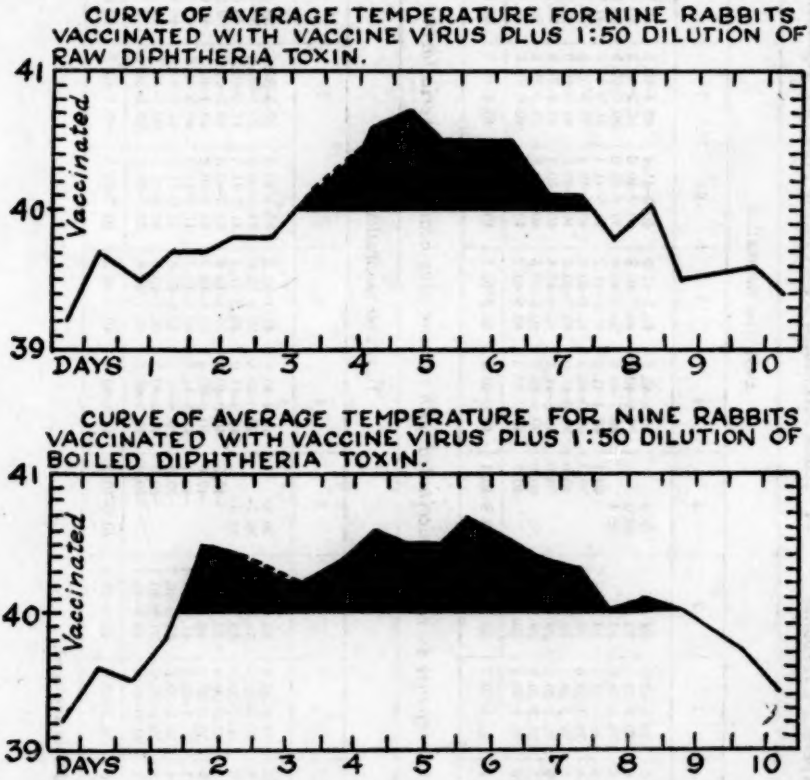
(*b*) *Temperature response in rabbits vaccinated with vaccine plus heated diphtheria toxin*

Rabbit No.	Days after vaccination										
	0	1	2	3	4	5	6	7	8	9	10
1580	39.2–39.3	39.2–39.5	40.8–	39.8–	40.4–40.8	41.1–41.3	40.5–40.0	39.8–39.9	39.5–40.0	40.2	
1582 *	38.6–39.5	39.0–39.5	39.8–	39.7–	39.6–39.8	39.5–39.0	39.5–39.9	39.5–39.8	39.4–39.5	39.0	
1584	39.1–40.0	39.7–39.9	40.7–	40.3–	40.9–41.0	40.2–41.4	41.2–41.3	41.0–40.8	40.0–40.4	40.7	
1610	39.2–39.5	39.0–39.4	40.8–40.8	–40.3	40.0–40.0	39.9–40.0	40.3–40.3	40.3–40.0	39.8–39.9	39.8	39.8–39.5
1612	39.8–39.9	39.6–39.5	40.0–40.2	–40.0	40.0–40.4	40.8–40.0	40.8–40.2	40.4–40.1	40.0–40.3	40.0	39.8–39.3
1614	39.0–39.5	39.1–40.0	41.0–40.4	–40.1	41.3–41.5	40.5–40.5	41.3–41.0	40.9–41.0	40.7–40.3	39.4	39.3–39.3
1616	39.5–39.8	39.3–39.8	40.0–40.2	–39.8	39.8–40.1	40.8–40.6	41.0–41.2	41.2–41.0	40.8–40.7	40.8	39.9–39.9
1618	39.6–39.8	39.2–39.7	39.8–40.1	–40.0	40.2–40.5	40.6–40.5	40.6–40.5	40.1–40.4	39.7–40.0	39.6	39.7–39.5
1620	39.1–39.4	41.0–40.8	40.9–40.7	–40.4	40.3–40.5	40.1–39.8	39.6–39.7	39.5–39.5	39.5–39.5	39.5	39.6–39.0
Average temperature	39.3–39.6	39.5–39.9	40.5–40.4	40.0–40.2	40.4–40.6	40.5–40.5	40.7–40.5	40.4–40.3	40.0–40.1	40.0	39.7–39.4

* 1582 proved to be immune to vaccinia (excluded from averages).

SYSTEMIC RESPONSE

Temperature.—Temperatures on experimental animals recorded twice daily are shown in Table 1. It will be noted that the temperatures in the virus-toxin group of nine animals tended to reach 40° C. from one to three days later and to return to normal earlier than was the case in the animals of the control group. The curves of average temperatures for the two groups are shown in the accompanying Chart.



Outcome.—Judged from general appearances, the animals vaccinated with the virus-toxin mixtures tended to withstand the resulting vaccinia better than did the controls. In order to test further this impression, a group of rabbits was vaccinated by a method which with virus 28628–88 would ordinarily cause some fatalities. Four rabbits were each vaccinated, in four areas one-half inch in diameter, with virus to which an equal volume of diphtheria toxin (1/50 dil.) had been added. Four control rabbits were similarly vaccinated, except that the toxin was previously heated for 15 minutes at 98° C. The

animals of the test group were all severely affected, but recovered, while three of the four control animals died (see Table 2). The autopsy findings were consistent with vaccinia in two of these, while in the third there was an accompanying consolidation involving parts of both lungs.

TABLE 2.—*The effect of adding diphtheria toxin to vaccine virus upon the mortality of intensively vaccinated rabbits*

Rabbit No.	Results		
	Local	General	Outcome
VACCINATED VIRUS+TOXIN ON NOVEMBER 11, 1931 (4 AREAS)			
1588	Modified	Moderate	Recovered.
1590	do	Severe	Do.
1592	do	do	Do.
1594	do	do	Do.
VACCINATED VIRUS+HEATED TOXIN ON NOVEMBER 11, 1931 (4 AREAS)			
1589	Very severe		Dead Nov. 18, 1931, of vaccinia.
1591	Severe		Do.
1593	Moderate	Severe	Recovered.
1595	Slight		Dead Nov. 17, 1931, of pneumonia+vaccinia.

HOW DOES DIPHTHERIA TOXIN EXERT ITS DETERRENT ACTION?

The fact that rabbits vaccinated with the virus-raw-toxin mixtures ran febrile courses, often with the development of secondary papules, together with the development of a subsequent immunity, proves that the virus was not killed by the toxin. Moreover, when rabbits which had been previously immunized against diphtheria, to the extent that they were Schick negative, were vaccinated with the virus-raw-toxin mixtures, no deterrent action was apparent. It was also found that when sufficient diphtheria antitoxin to prevent the "Schick" response was added to the virus-raw-toxin mixture the deterrent effect upon the "take" was also obliterated. It must be concluded, therefore, that the effect described is the result of the local cellular reaction engendered by the toxin at the site of the vaccination. This conclusion is supported by the fact that other agents which call forth a reaction may also exert a deterrent influence upon the local vaccine "take." For instance, it was found that a culture of N. Y. 5 scarlet fever streptococci, when added to vaccine virus, caused a definite deterrent action upon cutaneous vaccinations in rabbits, though less marked than was the case when diphtheria toxin was employed.

TABLE 3.—*Influence of Schick reaction upon subsequent vaccination in reaction area*

Rabbit No.	Date Schick tested	Date vaccinated	Interval in days between Schick test and vaccination	Vaccination results		Remarks
				Schick test area	New site	
1628	Dec 4, 1931	Dec. 5, 1931	1	+	+++	
1627	do	do	1	±	++++	
1626	do	Dec. 7, 1931	3	±	++++	
1625	do	do	3	+++	+++	
1524	do	do	3	−	++++	Dec. 17, 1931, dead of vaccinia.
1623	do	Dec. 9, 1931	5	++	++++	
1622	do	do	5	+++	++++	
1609	Nov. 17, 1931	Dec. 7, 1931	20	−	+++	
1608	do	do	20	++	++	
1606	do	do	20	±	++	

±=Questionable virus response.
+=Definite but slight take.
++=Slight necrosis, local edema.
+++=Moderate necrosis, marked local edema.
++++=Large necrosis with extensive massive edema.

Moreover, it was found that the site of a positive Schick response in rabbits remains relatively refractory to vaccine virus for at least 20 days, at which time the presence of toxin at the site seems improbable (see Table 3). A similar tendency was noted by Ledingham for areas previously injected with India ink or with cultures of *Streptococcus erysipelatis*.

SUMMARY

1. Diphtheria toxin when added in suitable amounts to vaccine virus exerts a deterrent local action upon the "take" in rabbits and renders the systemic response less severe.

2. The deterrent action of diphtheria toxin upon vaccinia is neutralizable by diphtheria antitoxin.

3. The deterrent action is apparently occasioned by the local cellular reaction rather than by any direct action of the diphtheria toxin upon the virus.

4. The site of a positive Schick response in rabbits remains relatively insusceptible to vaccine virus for at least 20 days.

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A STUDY OF SOME VITAMIN B ADSORBATES

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The use of fuller's earth or similar adsorbents for removing vitamin B from extracts of yeast or other raw materials, as proposed by Seidell (1), has often been resorted to as a means for concentrating this vitamin. An adsorbate of this type has been recommended as the international standard antineuritic vitamin preparation by the Conference on Vitamin Standards held in London in June, 1931 (2). More precise information in regard to the chemical character of such adsorbates is, therefore, much to be desired.

Although, when applied under proper conditions, fuller's earth is a very good adsorbent for the vitamin B complex, it always simultaneously adsorbs and carries down with it from the crude extract other substances besides the vitamin. The elimination of these co-adsorbed impurities at a later stage in processes for isolating vitamin B fractions is attended with difficulties and losses.

The underlying reason for the present study was the belief that a better knowledge of the nature, or of the properties, of these co-adsorbed impurities might conceivably lead to an improved method for their elimination. The same idea has recently been expressed by Van Veen (3), in connection with his studies on the isolation of the antineuritic vitamin from rice polishings. In our case, fresh bottom yeast from a near-by brewery was the source of the vitamin, and fuller's earth from Surrey, England, the adsorbent.

The immediate plan was to inquire to what extent adsorbates made from crude extracts of fresh brewers' yeast will contain, besides the adsorption compounds of vitamin B and other bases, loosely attached substances, and whether these and possibly other co-adsorbed impurities of the vitamin can be removed from the adsorbates by the use of inexpensive solvents without too great an impairment or loss of vitamin.

Very little information regarding these points can be found in the literature. A. de Cugnac (4) reported that the vitamin B fuller's earth adsorption complex is insoluble in formic acid. This acid, therefore, belongs to the group of liquids which, like water, fail to dissolve the vitamin when adsorbed on fuller's earth, although they are good solvents for the "free" vitamin. (This group of liquids does not include acetone, because in the absence of water acetone apparently

will not dissolve either "free" vitamin B or its adsorption compounds with fuller's earth.)

Narayanan and Drummond (5) studied the action of two acid and two alkaline liquids on a fuller's earth adsorbate of the pellagra-preventing factor of vitamin B from yeast. The adsorption compound was practically insoluble in the four liquids tried. The authors noted that the solvents in each case removed organic material; like De Cugnac, however, they were not interested in removing impurities, but only in finding a solvent for the adsorbed vitamin.

The present experiments were carried out with adsorbates prepared by two different methods. The two materials will be referred to for convenience as adsorbate P and adsorbate S. The latter was prepared from an aqueous extract of brewers' yeast in the manner described by Seidell (6), whereas the adsorbate P was made by treating pressed brewers' yeast essentially in the same manner which Pirie (7) has recently used for extracting the glutathione, then diluting the mixture with water, separating by means of a Sharples supercentrifuge, adding fuller's earth to the effluent, and washing and recovering the adsorbate.

Both adsorbate P and adsorbate S were hygroscopic gray powders which, as indicated in Table 3, contained nearly the same amounts of vitamin B.

The substances whose solvent action on the two adsorbates has been studied so far belong to three groups, namely:

(1) Neutral organic solvents of low boiling point, especially acetone.

(2) Mixtures of water and acetic acid varying in concentration from 50 to 100 per cent acid.

(3) A dilute aqueous solution of two mineral acids prepared by mixing equal parts of 1 per cent hydrochloric and 1 per cent hydrofluoric acid.

The latter mixture is referred to in this paper as Rather's reagent. Its use was decided upon after it had been observed that this mixture of dilute mineral acids, when applied to 1-gram portions of our fuller's earth preparations in the manner prescribed by Rather (8), would dissolve a large portion of the organic matter. (See second and third columns of figures in Tables 1A and 1B.)

The extraction experiments with low boiling solvents were carried out in Soxhlet extractors. One hundred grams of predried adsorbate was the quantity usually treated in each apparatus. The extractions were allowed to proceed for at least 9 or 10 days, during which time a calcium chloride tube was attached to the condensor of each apparatus. After an extraction was finished the residual adsorbate was freed from solvent by gentle heating, while the extract was filtered and evaporated in a tared dish or beaker. Volatile substances were driven off during this treatment, and, in weighing the dried residue, only the weight of the extracted nonvolatile matter which

had a dark brown, oily appearance and an acid reaction, was determined.

The first solvents tried were U.S.P. chloroform and acetone. The latter was dried with calcium chloride and redistilled before use. Adsorbate P which had been dried in a vacuum desiccator over sulphuric acid was used for these experiments. It was found by means of Seidell's preventive pigeon test (9) that through exhaustive extraction with either acetone or chloroform the adsorbates suffered no loss of vitamin. The amount of nonvolatile impurities removed was 0.75 grams from 100 grams of adsorbate P when using acetone and 0.10 grams when using U.S.P. chloroform. For the purpose of purifying an adsorbate of this type, acetone should, therefore, be given preference over U.S.P. chloroform.

Since, as shown in Table 1A, the total quantity of adsorbed organic matter contained in 100 grams of the original adsorbate P was 6.12 grams, the 0.75 grams of nonvolatile extract (assuming the latter to consist wholly of organic matter) would represent approximately 12 per cent of the adsorbed organic matter contained in adsorbate P. The higher proportion of acetone soluble (fatty) material in adsorbate P as compared with adsorbate S (see below) is doubtless due to the fact that fat solvents, namely, ethanol and ether, had been used in its preparation from pressed yeast in accordance with the method of Pirie (7). Seidell and Birckner (10) after attempting to extract and concentrate the vitamin B from an adsorbate of this type by a method which was known to be applicable to adsorbates prepared from aqueous extracts of yeast, reported that in this case their final product was unsatisfactory. It is possible that their failure was due in no small degree to the higher proportion of fatty impurities which were undoubtedly present in the "activated solid" prepared after treatment of the yeast with Pirie's reagent.

It was found that when acetone is brought in contact with a vitamin B adsorbate of the type here dealt with it not only acts as a solvent for loosely adsorbed substances but a portion of it is retained by the adsorbate. It may be inferred from the liberation of heat and the increase in weight that a chemical combination between acetone and fuller's earth has taken place. This is also indicated by the fact illustrated in Tables 1A and 1B that the acetone-treated adsorbates from which the free acetone had been wholly removed contained considerably more adsorbed organic matter than the untreated adsorbates (in spite of the fact that certain substances had been dissolved from the latter). The solvent effect of the acetone is, therefore, not apparent from the data in the second column of these tables.

In another experiment the adsorbate P prior to extraction with acetone had been heated in the drying oven at the temperature of

94° C. for about two days. The grey color of the adsorbate had thereby been changed to a tan. The amount of nonvolatile material removable by acetone from 100 grams of this heated adsorbate was only 0.6 grams, instead of 0.75 grams removed from the unheated adsorbate P. There are indications that on being heated the original adsorbate P not only undergoes oxidation but that some of its constituents are volatilized at the same time. To the latter factor may be due the lower yield of nonvolatile acetone-soluble extract above reported.

The results obtained by extracting adsorbate S with acetone were similar to those obtained in the case of adsorbate P. Prior to extraction the adsorbate S had been heated for several hours in the drying oven at 95° C. and finally for a few minutes at 110° C. The quantity of nonvolatile impurities removed with acetone from 100 grams of this adsorbate was 0.54 grams, which in this case was only about 5 per cent of the total adsorbed organic matter.

The treatments with acetic acid and with the mixture of dilute mineral acids were carried out by placing equal quantities of the respective adsorbate, which in these cases had been pre-extracted with acetone, into two 250-cubic centimeter Pyrex centrifuge bottles, adding about 150–200 cubic centimeters of the acid solvent and immersing the stoppered bottles in a water bath at 77–79° C. for one-half hour with frequent shakings. The mixtures were then centrifuged, the solutions poured off, and the residues stirred up with fresh solvent and digested and centrifuged in the same manner several times. The residues were finally freed from solvent, dried, and fed to pigeons as sole supplements to polished rice. The extracts were collected in volumetric flasks and aliquot portions were used for the analyses.

Immediately preceding the extractions with 70 and 90 per cent acetic acid, respectively, several extractions with 100 per cent acetic acid were made in the case of adsorbate P. Since, however, as indicated in Table 2, very little material was dissolved by the 100 per cent acid, these treatments were omitted in the case of adsorbate S.

It was thought that analytical data on the variously treated adsorbates and on the resulting extracts would be helpful in evaluating the relative efficacy of the different purification procedures studied. It was, therefore, decided to determine in some of these materials the percentage of nitrogen as well as that of total organic matter. For the latter purpose the method proposed by Rather (8) was tried, only to be abandoned when it was found that it gave results which were obviously much too low. The adsorbed organic matter was finally determined by igniting in a well-controlled electric muffle the carefully dried preparations and deducting from the loss on ignition the loss on ignition of the dried fuller's earth alone after it had been subjected to the same treatment as that employed in the preparation of

the respective adsorbate. The figures for organic matter in Tables 1A and 1B are, therefore, the losses on ignition as found, less the respective loss on ignition of the fuller's earth alone.

TABLE 1A.—*Changes in composition of adsorbate P due to treatment with acetone and to exposure to different temperatures*

Description of material	Total nitrogen by Kjeldahl method	Total adsorbed organic matter	Organic matter, insoluble in Rather's reagent
	Per cent	*Per cent*	*Per cent*
Original unheated material, dried in vacuo over P_2O_5	1.37	6.12	2.37
Original unheated material, defatted with acetone	1.30	9.53	1.87
Original material heated for 2 days at 62° C. and dried	1.44	6.42	2.90
Original material heated for 2 days at 94° C.	1.43	7.06	2.65
Material heated to 94° C. then defatted with acetone	1.40	9.40	1.99
Original material, dried at 100° C.	1.44	7.04	1.47

TABLE 1B.—*Changes in composition of adsorbate S due to heat-drying and to extraction with acetone and Rather's reagent*

Description of material	Total nitrogen by Kjeldahl method	Total adsorbed organic matter	Organic matter, insoluble in Rather's reagent
	Per cent	*Per cent*	*Per cent*
Original unheated material, dried in vacuo over P_2O_5	1.88	10.58	4.02
Original material, dried at 103° C.	1.90	10.92	2.22
Original material heated 4 hours at 95°, then 5 minutes at 110°, then defatted with acetone	1.74	12.84	4.45
Same material after additional treatment with Rather's reagent	1.80	11.68	(1)

[1] Not determined.

The above tabulations contain interesting information, yet the data are likely to be misleading unless they are carefully interpreted. From the nitrogen figures in conjunction with those for total adsorbed organic matter it follows with certainty only that for the original adsorbates P and S the nitrogen content of the organic matter was about 22 and 18 per cent, respectively. It would be incorrect to conclude that the treatment with Rather's reagent had removed no appreciable quantities of nitrogen or organic matter. As a matter of fact, Rather's mixture in acting upon the defatted adsorbate S had dissolved over one-third of the solid material. The precise record of this experiment was as follows:

Amount of material treated, 35.4 g., containing { 4.545 g. organic matter. / 0.616 g. nitrogen. / 319 vitamin (pigeon) units.

Amount of insoluble residue after 6 treatments with Rather's mixture and 3 washings with water, 22.4 g., containing { 2.616 g. organic matter. / 0.403 g. nitrogen. / 287 vitamin units.

Material (per cent) removed by the treatment: 36.72 for total solids, 42.44 for organic matter, 34.58 for nitrogen, 10 for vitamin B.

With reference to the above data and those in Tables 1A and 1B, it is therefore to be noted that it is not permissible to base calculations regarding nitrogen distribution solely on the nitrogen content of the solid residues as seems to have been done by Van Veen [1] (3).

In Tables 1A and 1B the gradual increase in the percentage of adsorbed organic matter with exposure of the adsorbates to higher temperatures is doubtless due to oxidation. The latter is a phenomenon of great prominence with these preparations, and it tends to overshadow other interesting properties.

While, as stated above, Rather's method for determining total organic matter was not applicable to our preparations, the results obtained with this method were nevertheless of very great interest. In the last columns of Tables 1A and 1B are, therefore, shown the percentages of organic material insoluble in Rather's mixture of dilute hydrochloric and hydrofluoric acids. The results are in this case not obscured and incumbered by oxidizable or oxidized materials, since apparently the bulk of these substances, together with a good many ingredients of the original fuller's earth, had been dissolved out. The acetone-fuller's earth adsorption compound, whose presence had caused abnormally high values for the total adsorbed organic matter, had also apparently been dissolved by the Rather reagent, with the result that in the last columns of Tables 1A and 1B the purifying effect of the acetone treatments is, at least in the case of adsorbate P, well demonstrated.

Above all, by applying the Rather method in conjunction with a reliable method for the estimation of the adsorbed organic matter, the fact was revealed that a large proportion of the organic ingredients of our fuller's earth preparations was soluble in Rather's reagent. Since, on the other hand, the fuller's earth adsorption compounds of the vitamin B fractions are known to be very resistant to the action of acids, it followed at once that Rather's mixture containing hydrofluoric acid, should be particularly well suited for purifying adsorbates of the type here dealt with. A preliminary experiment to study this point was, therefore, carried out with our defatted adsorbate S. The chemical changes effected have already been reported. The proportion of material treated with the acid mixture in this experiment was

[1] Van Veen also appears to be in error when he states that his acid clay adsorbate contained nearly 9 per cent of organic matter. Since the loss on ignition of his clay alone was 5 per cent and that of the adsorbate 10 per cent, the amount of adsorbed organic matter can only have been about 5 per cent. Hence, by the subsequent treatment with barium hydroxide Van Veen probably dissolved from one-half to two-thirds of the organic matter, instead of only one-fourth as stated in his paper.

considerably higher than that prescribed in Rather's analytical method, while the temperature during the digestions was only 77–79° C. It is perhaps due to these factors that, in the case of this experiment, only slightly over one-third of the adsorbed nitrogen and organic matter was dissolved by the acid mixture. A decrease, amounting to one-tenth of the total, in the vitamin B content of the residue had occurred in consequence of the digestions. The precise cause of this loss is not known at present. It is hoped, however, that by further experimentation with combinations of dilute mineral acids analogous to the one here employed a procedure may be developed for appreciably purifying previously defatted vitamin B adsorbates on fuller's earth without at the same time causing any appreciable loss of active substance.

The extraction of our two adsorbates with acetone has been discussed above. It should be added that as revealed by the odor during evaporation, the acetone extract contained also, at least in the case of adsorbate P, a certain undetermined amount of volatile ingredients. Since it followed from the experiments referred to that in the absence of water acetone will not dissolve the vitamin B adsorption compounds on fuller's earth, it seemed of interest to compare the behavior of this solvent toward the vitamin B as found in dried brewer's yeast. The following experiment with a commercial brand of dried brewer's yeast was, therefore, made:

A shallow layer of the ground yeast was placed in a vacuum desiccator over calcium chloride for three days. Eighty-three grams of the dry material were then extracted for a period of 13 days with dried, redistilled acetone, using a Soxhlet apparatus. The acetone dissolved fatty and resinous ingredients, and the undissolved residue, after evaporation of the solvent, was approximately 2.1 per cent lighter than the material at the start. Proportional amounts, namely, 0.39 and 0.40 grams per day, respectively, of the acetone-treated and of the untreated yeast were now fed to two groups of pigeons as the sole supplements to polished rice in the manner employed by Seidell (9). The average total change in weight of each pigeon during the 11-day feeding period was as follows:

For the birds receiving the untreated yeast..... −4.5 grams;
For the birds receiving the acetone-extracted yeast..... +1.0 gram.

It is, therefore, evident that through the prolonged extraction with acetone the yeast had not suffered any loss of vitamin B.

In the case of the acetic acid treatments of our two adsorbates, nitrogen determinations before and after extraction showed nothing of significance; and since the residues were obviously contaminated with various basic acetates (and possibly a fuller's earth adsorption compound of the acetic acid itself), it was deemed useless to attempt

a determination of the organic matter. In the case of adsorbate P some analytical determinations on the acetic acid extracts were made, however. These results which were obtained after driving off the free acetic acid, were as follows:

TABLE 2.—*Materials found in the acetic acid extracts of 100 grams of adsorbate P previously defatted with acetone*

Description of extract	Total solids		Ash		Nitrogen	
	Grams		*Grams*		*Grams*	
100 per cent acetic acid	0.37		0.11		0.0040	
100 per cent, followed by 90 per cent acetic acid	0.37	1.52	0.11	0.47	0.0040	0.0197
	1.15		0.36		0.0157	
100 per cent, followed by 70 per cent acetic acid	0.37	2.28	0.11	0.68	0.0040	0.0497
	1.91		0.57		0.0457	

A considerable, as yet undetermined, portion of these dissolved substances consisted not of co-adsorbed impurities, but of the acetates of inorganic bases removed from the fuller's earth by the hot acetic acid. For this reason only the nitrogen figures can be regarded as a criterion for the relative amounts of co-adsorbed impurities removed by these treatments. It is noteworthy that the 100 per cent acetic acid dissolved much less material than the 90 per cent and the 70 per cent acid. As is shown by the data in Table 3, the adsorbates, as a result of the treatments with acetic acid, had suffered no loss of vitamin B.

It remains to record briefly the feeding tests on pigeons with our variously treated fuller's earth preparations. These results are tabulated herewith:

TABLE 3.—*Changes in the body weights of pigeons which received as sole supplement to polished rice the variously treated adsorbates P and S*

Experiment No.	Description of vitamin supplement fed in addition to polished rice	Average daily dose	Number of pigeons used	Duration of test	Average total change in weight per pigeon
		Mg.		*Days*	*Grams*
I	No vitamin supplement		4	11	−40.8
II	Unheated adsorbate P	125	5	10	−0.4
	Unheated adsorbate P, exhaustively extracted with acetone.	125	3	10	+0.7
III	Unheated adsorbate P, extracted first with acetone, then with 100 per cent acetic acid.	130	3	14	+3.0
	Unheated adsorbate P, extracted in succession with acetone, 100 per cent, and 70 per cent acetic acid.	130	2	14	+5.0
	Unheated adsorbate P, extracted in succession with acetone, 100 per cent, and 90 per cent acetic acid.	130	2	14	+15.0
IV	Adsorbate P, dried at 94° C	130	3	11	−13.3
	Adsorbate P, dried at 94° C., then extracted with acetone.	130	3	11	−11.0
	Adsorbate P, dried at 94° C., then extracted with mixture of acetone and 100 per cent acetic acid.	130	3	11	−9.0
V	Adsorbate S	130	3	12	−5.0
	Adsorbate S, exhaustively extracted with acetone	130	4	12	−6.0
VI	Adsorbate S, exhaustively extracted with acetone	106	4	12	−8.0
	Adsorbate S, extracted first with acetone, then with 50 per cent acetic acid.	106	4	12	−10.5
	Adsorbate S, extracted first with acetone, then with 70 per cent acetic acid.	106	4	12	−6.8
VII	Adsorbate S, exhaustively extracted with acetone	111	5	10	+0.2
	Adsorbate S, extracted first with acetone, then with Rather's reagent.	111	5	10	−7.4
	Adsorbate S, extracted first with acetone, then with Rather's reagent.	123	4	10	−0.3

The pigeons which served for Experiments I to V belonged to a lot of 20 birds which had been on the basal diet of polished rice for several months at the time the present experiments were begun. Other pigeons were used for Experiments VI and VII. In each experiment the birds of the different groups were kept in the same large cage, and the comparative feeding tests with the different materials were carried out simultaneously.

In preparing the supplementary doses, the changes in weight which the adsorbates had undergone by being acted upon by the respective solvent were taken into account as far as possible. However, in the first column of figures in Table 3 the dosage is always expressed in terms of the respective untreated material.

It follows from Experiments II and IV that, in the case of adsorbate P, prolonged exposure to the temperature of 94° C. had caused some impairment of the vitamin B function.

SUMMARY

Vitamin B adsorbates on fuller's earth, when prepared from extracts of brewers' yeast, were found to contain impurities of a fatty nature. When these were removed from the dried adsorbates by means of acetone, it was found that the insoluble residue retained all of the vitamin B activity.

When for the original extraction of the yeast water alone had been used (adsorbate S), the proportion of the acetone-soluble nonvolatile impurities contained in the fuller's earth adsorbate was as low as 5 per cent of the total adsorbed organic matter. In a case where fat solvents had entered into the extraction of the yeast, the proportion of the acetone-soluble impurities was higher (10 to 12 per cent in our adsorbate P).

In making the extractions with acetone, a portion of the latter was retained by the adsorbates, and, consequently, the percentage of organic matter in the adsorbates, as well as their total weight, instead of being decreased by the removal of fatty matter, were increased by the treatments with acetone.

When the defatted adsorbates were treated with acetic acid of 70 to 100 per cent strength at a temperature of 77° to 79° C., a further small quantity of co-adsorbed impurities was dissolved without diminishing the vitamin potency of the insoluble residue.

A much larger portion of the co-adsorbed impurities was found to be removable from the defatted adsorbates by a mixture of equal volumes of 1 per cent hydrochloric and 1 per cent hydrofluoric acid (Rather's reagent). A 10 per cent decrease in the vitamin potency of the insoluble residue resulted from a preliminary trial with this treatment.

It is probable that extraction of vitamin B fuller's earth adsorbates with acetone and subsequently with a mixture of mineral acids similar to the one just referred to could be introduced as a useful intermediary step whenever the extraction of the vitamin from the adsorbate and the ultimate isolation of the active substance are being attempted.

A method has been indicated for the accurate determination of the adsorbed organic matter present in fuller's earth adsorbates of organic materials.

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DEATHS DURING WEEK ENDED DECEMBER 17, 1932

[From the Weekly Health Index, issued by the Bureau of the Census, Department of Commerce]

	Week ended Dec. 17, 1932	Corresponding week 1931
Data from 85 large cities of the United States:		
Total deaths	8,865	8,001
Deaths per 1,000 population, annual basis	12.7	11.6
Deaths under 1 year of age	646	581
Deaths under 1 year of age per 1,000 estimated live births [1]	53	46
Deaths per 1,000 population, annual basis, first 50 weeks of year	11.1	11.7
Data from industrial insurance companies:		
Policies in force	69,459,495	74,393,239
Number of death claims	13,769	13,691
Death claims per 1,000 policies in force, annual rate	10.4	9.6
Death claims per 1,000 policies, first 50 weeks of year, annual rate	9.5	9.6

[1] 1932, 81 cities; 1931, 77 cities.

PREVALENCE OF DISEASE

No health department, State or local, can effectively prevent or control disease without knowledge of when, where, and under what conditions cases are occurring

UNITED STATES

CURRENT WEEKLY STATE REPORTS

These reports are preliminary, and the figures are subject to change when later returns are received by the State health officers

Reports for Weeks Ended December 24, 1932, and December 26, 1931

Cases of certain communicable diseases reported by telegraph by State health officers for weeks ended December 24, 1932, and December 26, 1931

Division and State	Diphtheria		Influenza		Measles		Meningococcus meningitis	
	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931
New England States:								
Maine	----	17	2	3	2	698	0	0
New Hampshire	----	1	----	----	----	7	1	0
Vermont	1	----	----	----	1	88	0	0
Massachusetts	37	50	8	7	140	249	2	0
Rhode Island	2	5	2	----	----	473	0	0
Connecticut	15	8	24	5	18	58	1	1
Middle Atlantic States:								
New York	52	119	[1] 177	[1] 13	441	160	5	4
New Jersey	29	29	50	7	170	22	1	1
Pennsylvania	113	88	----	----	261	432	3	5
East North Central States:								
Ohio	30	101	47	2	341	103	0	2
Indiana	59	85	1,454	26	13	39	5	9
Illinois	73	95	336	----	42	29	14	3
Michigan	25	40	74	----	271	75	0	3
Wisconsin	7	8	492	23	409	24	0	0
West North Central States:								
Minnesota	9	19	45	1	271	14	1	1
Iowa	25	24	8	----	----	2	0	0
Missouri	15	56	384	6	----	3	3	4
North Dakota	6	10	----	----	131	----	1	0
South Dakota	3	5	208	----	----	33	0	0
Nebraska	16	20	941	----	18	25	1	1
Kansas	21	38	(?)	2	9	12	2	1
South Atlantic States:								
Delaware	----	11	3	----	1	1	0	0
Maryland [2]	18	61	353	22	3	10	0	0
District of Columbia	3	12	54	----	2	----	0	0
Virginia	11	----	----	----	92	----	3	----
West Virginia	24	41	517	8	150	258	0	1
North Carolina [4]	22	39	340	4	62	35	1	1
South Carolina	5	9	1,060	252	43	9	0	0
Georgia [4]	11	29	2,429	35	----	2	1	1
Florida	----	19	53	1	1	----	0	1

See footnotes at end of table.

Cases of certain communicable diseases reported by telegraph by State health officers for weeks ended December 24, 1932, and December 26, 1931—Continued

Division and State	Diphtheria		Influenza		Measles		Meningococcus meningitis	
	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931
East South Central States:								
Kentucky	29	42	1,004				7	0
Tennessee	22	52	2,945	31	6	8	0	5
Alabama [4]	22	43	3,965	21	1	28	1	0
Mississippi	9	36					1	0
West South Central States:								
Arkansas	12	19	9,795	7	4	11	1	1
Louisiana [4]	23	33	9,162	8		2	1	1
Oklahoma [4]	11	32	2,203	23		7	0	0
Texas	84	74	2,838	7	361	3	0	0
Mountain States:								
Montana	1	1	4,200		191	43	0	0
Idaho	3	2	2		2		0	0
Wyoming		2	243			3	0	0
Colorado	10	3	263		7	5	0	1
New Mexico	10	27	11	1		8	0	1
Arizona	1	6	33	3			0	0
Utah [3]	1		47	1	1	3	1	0
Pacific States:								
Washington	7	1	232		3	149	1	0
Oregon		2	1,552	34	39	9	0	0
California	39	60	1,068	79	48	43	4	2
Total	916	1,472	48,624	632	3,555	3,183	62	50

Division and State	Poliomyelitis		Scarlet fever		Smallpox		Typhoid fever	
	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931
New England States:								
Maine	1	0	31	18	0	0	4	2
New Hampshire	0	0	16	12	0	0	0	0
Vermont	0	0	2	10	0	25	0	1
Massachusetts	0	2	309	292	0	0	6	1
Rhode Island	0	0	11	25	0	0	0	0
Connecticut	1	1	83	57	13	12	2	1
Middle Atlantic States:								
New York	0	7	470	369	3	3	7	23
New Jersey	0	2	182	113	0	0	2	1
Pennsylvania	5	2	596	428	0	0	21	10
East North Central States:								
Ohio	1	2	236	419	17	6	2	10
Indiana	0	1	84	83	4	8	5	3
Illinois	1	2	390	257	1	36	4	19
Michigan	0	4	337	246	1	7	8	8
Wisconsin	0	1	76	106	3	13	0	0
West North Central States:								
Minnesota	0	1	70	75	0	6	0	1
Iowa	0	1	40	35	19	48	1	5
Missouri	0	1	24	55	0	14	2	6
North Dakota	0	0	7	17	5	11	0	0
South Dakota	0	0	14	7	2	3	0	6
Nebraska	1	0	40	19	2	12	0	0
Kansas	4	0	73	61	2	8	5	1
South Atlantic States:								
Delaware	0	0	10	6	0	0	0	1
Maryland [3]	0	0	94	74	0	0	7	9
District of Columbia	0	0	10	8	0	0	0	1
Virginia	0	1	52		1		6	
West Virginia	0	1	61	49	0	2	7	14
North Carolina [4]	0	2	60	49	1	0	2	3
South Carolina	0	0	5	7	0	0	0	8
Georgia [4]	1	0	9	7	0	0	3	5
Florida	0	1	8	0	0	1	1	0

See footnotes at end of table.

Cases of certain communicable diseases reported by telegraph by State health officers for weeks ended December 24, 1932, and December 26, 1931—Continued

Division and State	Poliomyelitis		Scarlet fever		Smallpox		Typhoid fever	
	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931	Week ended Dec. 24, 1932	Week ended Dec. 26, 1931
East South Central States:								
Kentucky	1	0	23	66	2	0	10	9
Tennessee	0	1	26	63	1	9	4	16
Alabama [4]	0	0	28	35	1	1	0	16
Mississippi	0	0	7	14	0	6	1	3
West South Central States:								
Arkansas	0	0	11	27	0	7	2	5
Louisiana [4]	0	0	7	12	8	0	3	39
Oklahoma [5]	0	0	26	56	3	1	0	8
Texas	0	1	78	58	6	9	2	12
Mountain States:								
Montana	1	1	8	35	0	13	1	1
Idaho	1	0	2	3	2	0	1	0
Wyoming	0	0	3	2	0	0	0	0
Colorado	0	0	28	23	0	11	0	0
New Mexico	0	1	11	17	0	0	1	10
Arizona	0	1	3	6	0	0	0	1
Utah [3]	0	0	19	7	0	0	0	0
Pacific States:								
Washington	2	1	37	35	6	45	3	1
Oregon	0	1	16	11	0	5	1	2
California	2	1	134	90	4	1	6	3
Total	22	40	3,865	3,464	107	323	130	265

[1] New York City only.
[2] The department of health of Kansas, after circularizing local health officers in the State, estimated that there were 78,624 cases of influenza in Kansas.
[3] Week ended Friday.
[4] Typhus fever, week ended Dec. 24, 1932, 7 cases: 1 case in North Carolina, 3 cases in Georgia, 2 cases in Alabama, and 1 case in Louisiana.
[5] Figures for 1932 are exclusive of Oklahoma City and Tulsa.

SUMMARY OF MONTHLY REPORTS FROM STATES

The following summary of cases reported monthly by States is published weekly and covers only those States from which reports are received during the current week:

State	Meningococcus meningitis	Diphtheria	Influenza	Malaria	Measles	Pellagra	Poliomyelitis	Scarlet fever	Smallpox	Typhoid fever
September, 1932										
Massachusetts	7	70	6	1	94	1	12	363	0	30
October, 1932										
New Hampshire		3					0	61	0	
November, 1932										
Georgia		266	309	385	3	20	1	103	0	55
Illinois	44	424	150	6	223		18	1,496	2	71
Louisiana	3	162	1,583	72	21	11	3	99	5	28
New Hampshire		1					0	81	0	
New Mexico	21	86	753		4		0	43	1	18
Ohio	8	329	492		582		6	2,096	111	58
Rhode Island	2	23			5		0	147	0	3
South Dakota	1	24	6		8		1	45	4	5
West Virginia	3	179	117		241		2	285	0	63

September, 1932

Massachusetts:	Cases
Chicken pox	81
Dysentery	4
German measles	34
Lead poisoning	2
Lethargic encephalitis	2
Mumps	111
Ophthalmia neonatorum	46
Septic sore throat	9
Tetanus	2
Trachoma	2
Trichinosis	1
Whooping cough	303

November, 1932

Chicken pox:	
Georgia	27
Illinois	2,003
Louisiana	36
New Mexico	68
Ohio	2,662
Rhode Island	63
South Dakota	182
West Virginia	258
Conjunctivitis:	
New Mexico	1
Dysentery:	
Georgia	7
Illinois (bacillary)	7
Louisiana	2
Ohio	3
Food poisoning:	
New Mexico	3
Ohio	14
German measles:	**Cases**
Illinois	18
New Mexico	2
Ohio	16
Rhode Island	1
Hookworm disease:	
Louisiana	17
Lead poisoning:	
Illinois	1
Ohio	19
Leprosy:	
Louisiana	2
Lethargic encephalitis:	
Georgia	1
Illinois	8
Mumps:	
Georgia	16
Illinois	139
Louisiana	1
New Mexico	12
Ohio	125
Rhode Island	16
South Dakota	11
West Virginia	1
Ophthalmia neonatorum:	
Illinois	5
Ohio	84
Paratyphoid fever:	
Illinois	1
Puerperal septicemia:	
Illinois	2
New Mexico	1
Ohio	6
Rabies in animals:	
Illinois	2
Louisiana	3
Septic sore throat:	**Cases**
Georgia	26
Illinois	16
Ohio	191
South Dakota	1
Tetanus:	
Illinois	3
Louisiana	10
Ohio	2
South Dakota	3
Trachoma:	
Illinois	3
Louisiana	10
Ohio	6
South Dakota	4
Trichinosis:	
Illinois	1
Tularaemia:	
Illinois	13
Ohio	11
Typhus fever:	
Georgia	30
Louisiana	1
Undulant fever:	
Illinois	8
Louisiana	5
New Mexico	1
Ohio	6
Vincent's angina:	
Illinois	23
Whooping cough:	
Georgia	44
Illinois	331
Louisiana	9
New Mexico	15
Ohio	328
Rhode Island	64
South Dakota	18
West Virginia	81

WEEKLY REPORTS FROM CITIES

City reports for week ended December 17, 1932

State and city	Diphtheria cases	Influenza cases	Influenza deaths	Measles cases	Pneumonia deaths	Scarlet fever cases	Smallpox cases	Tuberculosis deaths	Typhoid fever cases	Whooping cough cases	Deaths, all causes
Maine:											
Portland	1	----	0	0	0	6	0	2	0	7	28
New Hampshire:											
Concord	0	----	0	0	1	0	0	0	0	0	9
Manchester	0	----	0	0	0	0	0	1	0	0	11
Nashua	0	----	0	0	0	1	0	0	0	0	0
Vermont:											
Barre	0	----	0	0	0	0	0	2	0	0	6
Massachusetts:											
Boston	19	3	1	45	36	80	0	9	1	60	240
Fall River	0	----	0	0	0	3	0	1	0	0	22
Springfield	0	1	0	0	7	9	0	1	0	2	38
Worcester	0	----	0	1	7	30	0	1	0	0	55
Rhode Island:											
Pawtucket	0	----	0	0	0	0	0	0	0	0	17
Providence	2	1	0	0	3	22	0	2	0	20	64
Connecticut:											
Bridgeport	0	----	0	6	4	5	0	1	0	9	27
Hartford	1	1	0	5	3	6	0	0	1	2	41
New Haven	0	----	0	0	4	3	0	1	0	6	31
New York:											
Buffalo	3	7	2	3	20	31	0	5	0	25	136
New York	43	45	14	265	170	184	0	76	3	110	1,467
Rochester	1	2	0	1	4	22	0	2	1	5	59
Syracuse	0	----	0	4	4	10	0	2	0	2	48
New Jersey:											
Camden	2	----	0	0	1	4	0	1	0	2	22
Newark	5	12	0	49	7	21	0	5	0	9	94
Trenton	3	4	0	1	5	9	0	1	0	2	39
Pennsylvania:											
Philadelphia	10	8	6	17	43	114	0	28	0	4	439
Pittsburgh	5	82	33	1	68	48	0	12	0	9	274
Reading	1	----	0	4	7	3	0	1	0	1	26
Scranton	1	----	----	1	----	7	0	----	0	2	----

City reports for week ended December 17, 1932—Continued

State and city	Diphtheria cases	Influenza		Measles cases	Pneumonia deaths	Scarlet fever cases	Smallpox cases	Tuberculosis deaths	Typhoid fever cases	Whooping cough cases	Deaths, all causes
		Cases	Deaths								
Ohio:											
Cincinnati	3	7	8	0	14	22	0	4	0	1	142
Cleveland	7	281	10	3	25	71	0	8	0	16	206
Columbus	3	17	11	75	14	10	0	4	0	1	120
Toledo	3	8	1	11	8	34	0	3	0	0	82
Indiana:											
Fort Wayne	9	------	1	0	1	3	0	0	0	0	26
Indianapolis	6	------	3	8	22	9	0	2	3	4	------
South Bend	0	------	1	0	1	3	0	0	0	1	17
Terre Haute	0	------	0	1	5	4	0	1	0	0	16
Illinois:											
Chicago	11	71	20	36	76	175	0	35	1	12	803
Springfield	1	9	0	0	4	4	0	1	0	0	22
Michigan:											
Detroit	24	32	4	58	27	105	0	25	1	55	242
Flint	1	24	1	2	5	6	0	0	0	4	31
Grand Rapids	1	------	4	1	3	10	0	0	0	27	------
Wisconsin:											
Kenosha	0	1	1	0	0	2	2	0	0	1	9
Madison	0	------	------	5	------	3	0	------	0	8	------
Milwaukee	4	5	4	2	14	11	0	7	0	14	110
Racine	0	------	0	0	2	10	0	0	0	1	13
Superior	0	------	0	0	0	0	0	0	0	7	9
Minnesota:											
Duluth	0	------	3	0	4	3	0	0	0	5	42
Minneapolis	1	4	9	18	17	17	0	0	0	4	105
St. Paul	0	2	2	2	5	18	0	2	0	23	80
Iowa:											
Des Moines	7	------	------	0	------	6	0	------	0	0	46
Sioux City	2	------	------	1	------	3	0	------	0	0	1
Waterloo	0	------	------	0	------	0	0	------	0	0	------
Missouri:											
Kansas City	2	2	4	14	35	35	0	8	0	0	86
St. Joseph	1	------	2	0	15	4	0	1	0	0	39
St. Louis	15	10	5	0	13	15	0	11	1	1	249
North Dakota:											
Fargo	0	------	0	0	1	0	0	[1]2	0	0	8
Grand Forks	0	------	0	36	0	0	0	0	0	0	0
South Dakota:											
Aberdeen	0	------	0	0	0	2	0	0	0	0	------
Nebraska:											
Omaha	8	------	0	0	16	9	1	2	0	0	74
Kansas:											
Topeka	2	------	0	2	4	3	0	0	0	2	6
Wichita	0	------	1	0	11	4	0	2	0	0	43
Delaware:											
Wilmington	1	------	0	1	2	3	0	0	0	0	23
Maryland:											
Baltimore	9	59	1	2	29	60	0	13	2	12	224
Cumberland	0	1	0	0	0	1	0	0	0	0	14
Frederick	1	------	0	0	0	3	0	0	0	0	5
District of Col.:											
Washington	5	64	4	0	23	12	0	10	0	5	175
Virginia:											
Lynchburg	1	------	0	1	1	1	0	0	0	0	13
Norfolk	0	------	0	0	4	0	0	3	0	0	27
Richmond	4	------	4	1	3	3	0	2	0	0	59
Roanoke	2	------	0	0	1	1	0	0	1	0	16
West Virginia:											
Charleston	0	15	2	0	3	3	0	1	0	0	17
Huntington	3	------	------	14	------	2	0	------	0	0	------
Wheeling	0	------	0	67	7	2	0	0	0	9	18
North Carolina:											
Raleigh	0	------	0	0	4	1	0	1	0	0	22
Wilmington	------	------	------	------	------	------	------	------	------	------	------
Winston-Salem	1	------	0	0	2	3	0	1	0	0	13
South Carolina:											
Charleston	1	66	3	0	1	2	0	5	0	0	31
Columbia	2	------	0	0	1	0	0	0	0	0	11
Greenville	0	------	0	2	0	0	0	0	0	0	------
Georgia:											
Atlanta	11	1,442	20	0	7	3	0	0	0	7	87
Brunswick	0	------	0	0	0	0	0	0	0	0	4
Savannah	0	81	3	0	2	3	0	1	1	0	22
Florida:											
Miami	2	1	0	0	4	0	0	2	0	0	22
Tampa	4	2	1	0	1	1	0	0	0	1	11

[1] Nonresidents.

City reports for week ended December 17, 1932—Continued

State and city	Diphtheria cases	Influenza		Measles cases	Pneumonia deaths	Scarlet fever cases	Smallpox cases	Tuberculosis deaths	Typhoid fever cases	Whooping cough cases	Deaths, all causes
		Cases	Deaths								
Kentucky:											
Covington											
Lexington	0	136	1	0	2	2	0	0	1	1	30
Louisville	8	74	2	0	20	7	0	3	0	3	106
Tennessee:											
Memphis	4		9	3	9	5	0	6	0	0	94
Nashville											
Alabama:											
Birmingham	4	884	18	0	10	7	0	4	0	0	104
Mobile	1	50	7	0	4	1	1	0	0	0	34
Montgomery	0	94		0		0	0		0	0	
Arkansas:											
Fort Smith											
Little Rock	0	1	0	0	0	2	0	0	0	0	
Louisiana:											
New Orleans	5	229	34	0	24	3	0	13	1	3	176
Shreveport	1		0	0	5	0	0	2	0	0	23
Oklahoma:											
Muskogee	1	13		0		3	2		0	0	
Tulsa	0			0		3	0		0	0	
Texas:											
Dallas	19	80	9	1	6	13	0	3	1	1	76
Fort Worth	6		5	0	6	14	0	1	0	0	36
Galveston	2		0	0	3	0	0	2	0	0	15
Houston	14		3	2	21	3	0	7	0	0	79
San Antonio	8	11	5	0	15	1	0	10	0	0	87
Montana:											
Billings	0		0	1	0	0	0	0	0	0	8
Great Falls	0		0	366	3	1	0	0	0	1	8
Helena	0	294	2	0	0	0	0	0	0	0	8
Missoula	0		0	0	0	1	0	0	0	0	6
Idaho:											
Boise	0		0	6	1	0	6	1	0	0	7
Colorado:											
Denver	3	307	12	7	32	13	0	4	0	1	112
Pueblo	0		1	0	2	0	0	1	0	0	10
New Mexico:											
Albuquerque	2	1	0	0	1	4	0	4	0	0	12
Arizona:											
Phoenix	0		0	0	3	0	0	4	0	0	
Utah:											
Salt Lake City	0		7	0	6	3	0	0	0	0	39
Nevada:											
Reno	0		0	0	0	0	0	0	0	0	1
Washington:											
Seattle	0			1		6	1		0	8	
Spokane	0			1		4	1		0	0	
Tacoma	0		1	0	1	2	0	1	0	0	25
Oregon:											
Portland	1	16	1	3	3	9	7	2	1	1	93
Salem	0	40		2		0	0		0	0	
California:											
Los Angeles	15	240	12	6	28	29	1	24	0	17	309
Sacramento	0	6	1	0	7	2	0	0	1	1	
San Francisco	2	126	10	2	16	3	0	15	1	26	213

City reports for week ended December 17, 1932—Continued

State and city	Meningococcus meningitis		Poliomyelitis cases
	Cases	Deaths	
Massachusetts:			
Boston	0	1	0
New York:			
Buffalo	1	0	0
New Jersey:			
Newark	0	0	1
Pennsylvania:			
Philadelphia	1	2	2
Pittsburgh	0	1	0
Ohio:			
Cleveland	0	0	1
Indiana:			
Indianapolis	2	2	0
Illinois:			
Chicago	7	3	0
Springfield	0	1	0
Michigan:			
Detroit	0	1	0
Minnesota:			
Duluth	2	0	0
Minneapolis	1	0	0
Missouri:			
Kansas City	3	2	0
Kansas:			
Topeka	2	2	0
District of Columbia:			
Washington	3	1	0
Kentucky:			
Louisville	0	1	0
Louisiana:			
New Orleans	2	0	0
Montana:			
Missoula	1	0	0
California:			
Los Angeles	0	2	2
Sacramento	1	0	0

Lethargic encephalitis.—Cases: New York, 2; Detroit, 2; Birmingham, 1.
Pellagra.—Cases: Baltimore, 1; Charleston, S. C., 1; Dallas, 1.
Typhus fever.—Cases: Charleston, S. C., 1; Tampa, 1.

FOREIGN AND INSULAR

CANADA

Provinces — Communicable diseases — Week ended December 10, 1932.—The Department of Pensions and National Health of Canada reports cases of certain communicable diseases for the week ended December 10, 1932, as follows:

Disease	Prince Edward Island	Nova Scotia	New Brunswick	Quebec	Ontario	Manitoba	Saskatchewan	Alberta	British Columbia	Total
Cerebrospinal meningitis					1					1
Chicken pox		37		111	370	98	36	10	26	688
Diphtheria	2	2	5	33	19	4		3		68
Erysipelas				3		2			1	6
Influenza		11			540		6		534	1,091
Lethargic encephalitis					1					1
Measles		7	14	57	523	4	3	31	7	646
Mumps		24			115	30			16	185
Pneumonia, all forms		4			18				11	33
Scarlet fever		20	16	84	82	33	14	16	15	280
Smallpox					3		2			5
Trachoma						2			2	4
Tuberculosis		2	5	88	23	16	19		11	164
Typhoid fever	1		2	18	3	5	1			30
Undulant fever					3					3
Whooping cough		1		77	64	38	7	1	45	233

CUBA

Provinces—Communicable diseases—Four weeks ended November 12, 1932.—During the four weeks ended November 12, 1932, cases of certain communicable diseases were reported in the provinces of Cuba as follows:

Disease	Pinar del Rio	Habana	Matanzas	Santa Clara	Camaguey	Oriente	Total
Diphtheria		17	3	3	3		26
Malaria	100	33	22	90	80	19	344
Measles			3		2	11	16
Scarlet fever		3	3				6
Tuberculosis		13	2	10	2	22	49
Typhoid fever	3	22	4	4	6	7	46

CZECHOSLOVAKIA

Communicable diseases—August–October, 1932.—During the months of August, September, and October, 1932, certain communicable diseases were reported in Czechoslovakia as follows:

Disease	August		September		October	
	Cases	Deaths	Cases	Deaths	Cases	Deaths
Anthrax	13	4	10		12	2
Cerebrospinal meningitis	7	5	6	3	7	3
Diphtheria	1,476	72	2,797	120	4,582	176
Dysentery	98	7	189	35	357	67
Malaria	66	1	58		34	1
Paratyphoid fever	32	1	28	1	13	2
Puerperal fever	49	25	27	11	45	10
Rabies	1	1				
Scarlet fever	1,160	17	1,940	12	3,302	28
Trachoma	161		149		181	
Typhoid fever	897	63	1,055	62	1,359	85
Typhus fever					6	2

YUGOSLAVIA

Communicable diseases—November, 1932.—During the month of November, 1932, certain communicable diseases were reported in Yugoslavia as follows:

Disease	Cases	Deaths	Disease	Cases	Deaths
Anthrax	83	21	Poliomyelitis	24	3
Cerebrospinal meningitis	5	3	Scarlet fever	544	16
Diphtheria and croup	1,966	247	Sepsis	14	5
Dysentery	315	57	Tetanus	29	12
Erysipelas	196	9	Typhoid fever	1,697	172
Measles	263	7	Typhus fever	3	1
Paratyphoid fever	110				

CHOLERA, PLAGUE, SMALLPOX, TYPHUS FEVER, AND YELLOW FEVER

(NOTE.—A table giving current information of the world prevalence of the quarantinable diseases appeared in the Public Health Reports for December 30, 1932, pp. 2382–2394. A similar cumulative table will appear in the Public Health Reports to be issued January 27, 1933, and thereafter, at least for the time being, in the issue published on the last Friday of each month.)

Cholera

Philippine Islands.—During the week ended December 24, 1932, cholera was reported in the Philippine Islands as follows: Leyte Province, 17 cases, 14 deaths; Samar Province, 9 cases, 4 deaths.

Plague

Argentina—San Luis Province.—On December 9, 1932, several cases of bubonic plague, one fatal, were reported in the town of Quines, San Luis Province, Argentina.

France—Marseille.—A case of plague was reported December 27, 1932, at Marseille, France.

Hawaii Territory—Plague-infected rats.—Plague-infected rats have been reported at Paauilo, Hamakua Territory, Island of Hawaii, as follows: December 16, 1 rat; December 17, 7 rats; December 25, 1 rat; December 26, 1 rat. Paauilo is in the interior, about 175 miles from Honolulu, which is on the island of Oahu. On December 24 a plague-infected rat was reported from Manienie Gulch, 1 mile from the location where the above-mentioned rats were found.

Yellow Fever

French West Africa—Guinea.—A fatal case of suspected yellow fever was reported December 7, 1932, at Koliagbe, Kindia Circle, Guinea, French West Africa.